

## Limited influence of hospital wastewater on the microbiome and resistome of wastewater in a community sewerage system

Buelow, Elena; Bayjanov, Jumamurat R; Majoor, Eline; Willems, Rob J L; Bonten, Marc J M; Schmitt, Heike; van Schaik, Willem

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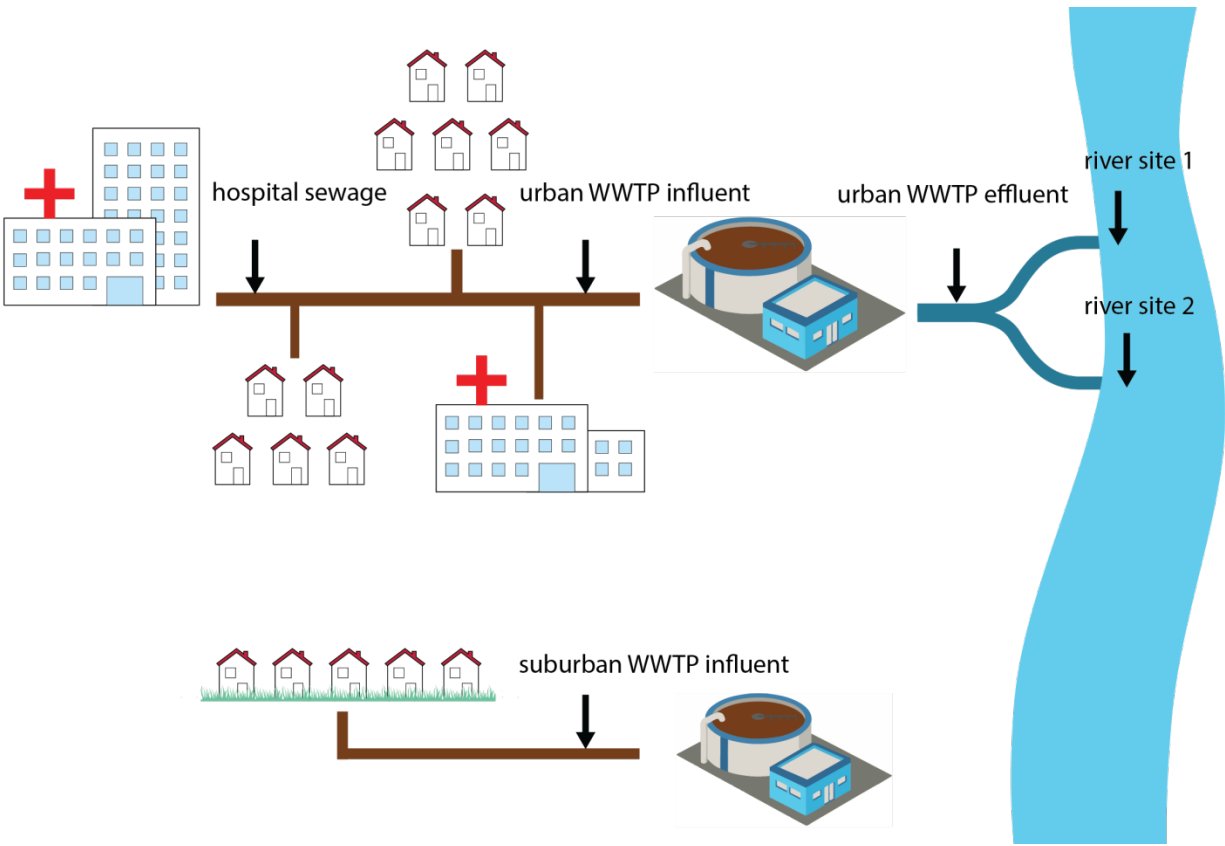
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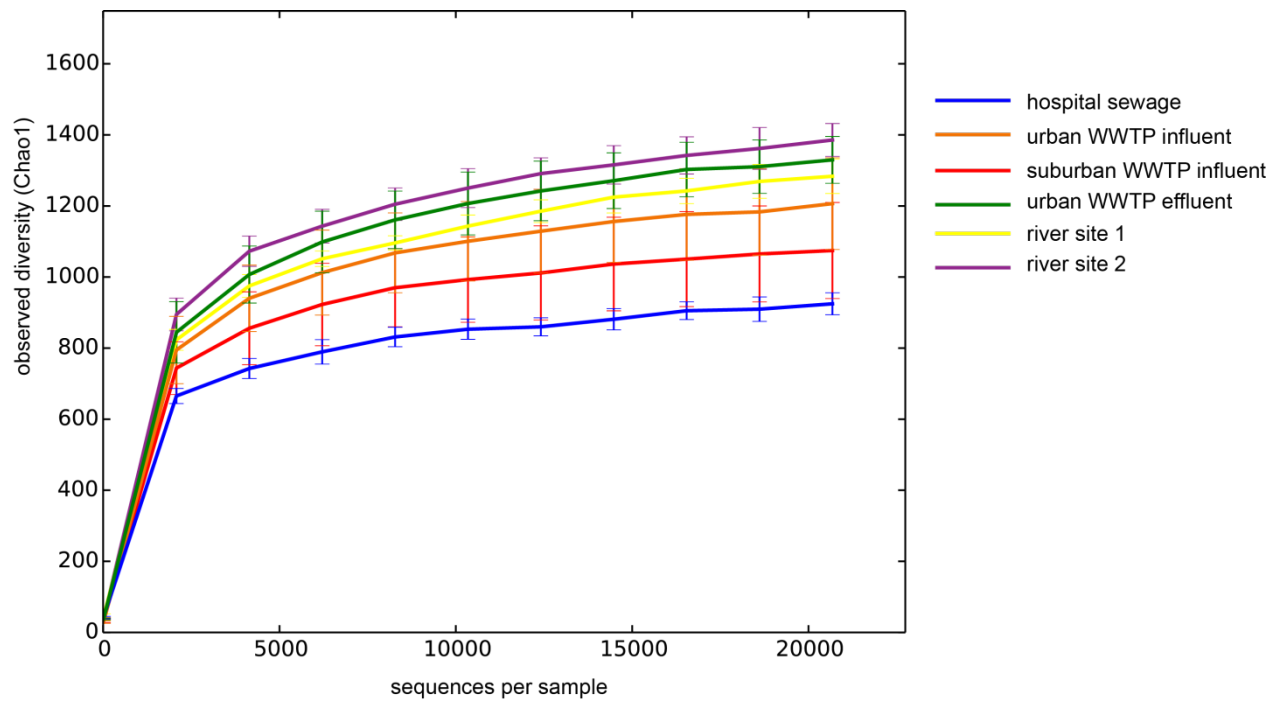
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**Supplementary Figures and Tables**



**Supplementary Figure 1: Schematic representation of sampling locations.** Sampling was carried out on four different days within the course of 2.5 weeks (Monday 31 March 2014; Wednesday 2 April 2014; Monday 7 April 2014; Monday 14 April 2014). The different sampling locations are indicated by black arrows. The vector graph depicting the sewage treatment plant was provided by Tracey Saxby, Integration and Application Network, University of Maryland Center for Environmental Science.



**Supplementary Figure 2: Observed diversity for the different sampling sites.** Rarefaction curves depict the Chao1 index which indicates the species diversity per sampling site as a function of the sub-sampling depth with the maximum threshold of 20681 sequences per sample.

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21 **Supplementary Table 1. Primers used in this study.** Primers were developed to target the indicated  
 22 ARGs. Primer sequences in bold indicate ARGs which were detected in  $\geq 1$  sample. Grey background  
 23 indicates modifications to primer sequences compared to those published in Buelow *et al.*, 2017.

Antimicrobial resistance gene	Accession number	Forward primer	Reverse primer
<b>catB3</b>	<b>ABD58915.1</b>	<b>AAGGCAAGCTGCTTTCTGAG</b>	<b>TCGAATGAGTGCCCATGATA</b>
<i>acrF</i>	CAR04877.2	ACTGACACCGGTTGATGTGA	GAGCAATAATCGAGGCGTTC
<i>tolC</i>	BAG78840.1	CTGAAAGAAGCCGAAAAACG	CGTCGGTAAGTGACCATCCT
<i>acrA</i>	ACI36997.1	GAAGGTAGCGACATCGAAGC	CTTTCGCCAGATCACCTTTC
<i>aph(3')-III</i>	ACB90577.1	CCGGTATAAAGGGACCACCT	CTTTGGAACAGGCAGCTTTC
<i>aph(2'')-Ib</i>	AF207840.1	ATCAAATCCCTGCGGTAGTG	CAAGGGCATCCTTTTCCTTT
<i>aadE</i> -like gene	AAW34138.1	GCATGATTTCTGGCTGATT	CCACAATTCTCTGGGACAT
<i>aac(6')-aph(2'')</i>	ABY79711.1	TCCAAGAGCAATAAGGGCATA	TGCCCTCGTGTAATTCATGT
<i>aac(6')-li</i>	WP_002293989.1	AGACAGCTCGGCAGAAGAAG	ACCGTATTGAGGGATTGCAC
<i>aac(3')-li(acde)</i>	HQ246166.1	TGACGTATGAGATGCCGATG	GAGAATGCCGTTTGAATCGT
<i>aac(6')-Ib</i>	KM387722.1	TTGCAATGCTGAATGGAGAG	TGGTCTATTCCCGCTACTCC
<b>aadA</b>	<b>ADW23165.1</b>	<b>CAGCGGAGGAATTCTTTGAC</b>	<b>GCTGCGAGTTCCATAGCTTC</b>
<b>aac(6')-IIa</b>	<b>ACR24243.1</b>	<b>GGAACACTACCTGCCAGAG</b>	<b>GCGACGTACGACTGAGCATA</b>
<i>aph(2'')-I(de)</i>	AAC14693.1	CGGAGGTGGTTTTACAGGA	TTGCTTCGGCAGATTATTGA
<i>aph(3')-Ia, -Ic</i>	CAQ58482.1	ATTCTCACCGGATTCACTCG	GATTCCGACTCGTCCAACAT
<i>strB</i>	CAJ77026.1	GGCGATTATAGCCGATCAAA	CGCGACTGGAGAACATGATA
<i>bacA_2</i>	ABR38862.1	GAGGCATTGATCCTTGGTGT	AAACAATGCCGAACCGATAG
<i>bacA_1</i>	CAH05846.1	GGCTGCGTTACTGTCTGTTT	GGCCAATGATAAATGCATCC
<i>bacA</i>	ACL18936.1	AACTTCCCGTTCTGGTGCTA	CATAACGGGGATAGCGAGAA
<b>bla<sub>GES</sub></b>	<b>ABG47465.1</b>	<b>GACCCACACCATTGAGAGGT</b>	<b>GCGCAGGTACCAGTTTTCTC</b>
<i>bla<sub>IMP</sub></i>	AJ640197.1	GCTACCGCAGCAGAGTCTTT	CCCACCCGTTAACTTCTTCA
<i>bla<sub>VIM</sub></i>	AM183120.1	TGTCCGTGATGGTGATGAGT	TTTCAATCTCCGCGAGAAGT
<i>bla<sub>ACC</sub></i>	AJ870923.1	TTGTTACGCTACGTGCAAGC	CGATTTGAAATAGCCGGTGT
<i>bla<sub>DHA</sub></i>	AHN96243.1	AAAGTGCGCAAAGCCAGTAT	AAGATTCCGCATCAAGCTGT
<i>bla<sub>IMI</sub></i>	U50278.1	AGTCGATCCCAGCAGCTTTA	CCAAGAACTGTGCATTCCA
<i>bla<sub>CMY-1/MOX</sub></i>	AF357598.1	GATCTGTGTCGTTTTGTGAA	CTACCGAGTAATGCCCTTGG
<i>bla<sub>AMPc</sub></i>	ABF06289.1	ACCGCTAAACAGTGGAATGG	GCAAGTCGCTTGAGGATTTTC
<i>cepA</i>	CR626927.1	ATGTCTCTGCCCTGGTAGTTG	CTTGCCCGTCGATAATGACT
<i>cepA_2</i>	AE016945.1	TGCACCAAGACGAAAGTCTG	ACAGTGCTTCTTTGCGGAAT
<i>bla<sub>BIC-1</sub></i>	GQ260093.1	CCATCAGCGCACACATAGT	CCAGAACGTTTTCCAGAAGC
<i>cbIA</i>	AAA66962.1	TGCCTGCGACATCTTGATAG	CCGTCTTCTGTTCCGAGAG
<i>cfxA</i>	AY769933.1	GCGCAAATCCTCCTTTAACA	ACAATAACCGCCACACCAAT
<i>bla<sub>CMY-2</sub></i>	AAZ99133.1	CGATCCGGTACGAAATACT	CCTGCCGTATAGGTGGCTAA
<i>bla<sub>CTX-M</sub></i>	ABG46354.1	ACTATGGCACCACCAACGAT	GGTTGAGGCTGGGTGAAGTA
<i>bla<sub>TEM</sub></i>	NP_775035.1	AAGCCATACCAAACGACGAG	TTGCCGGGAAGCTAGAGTAA
<b>bla<sub>SHV</sub></b>	<b>AAV83796.1</b>	<b>CTTTCCCATGATGAGCACCT</b>	<b>CAGATCTGCTGGCGATAGT</b>
<b>bla<sub>NDM</sub></b>	<b>CAZ39946.1</b>	<b>TGGATCAAGCAGGAGATCAA</b>	<b>ATTGGCATAAGTCGCAATCC</b>
<i>bla<sub>OXA</sub></i>	AAP70012.1	GTGGCATCGATTATCGGAAT	AGAGCACAACTACGCCCTGT
<i>bla<sub>KPC</sub></i>	AEL12451.1	TGGCTAAAGGGAAACACGAC	TAGTCATTTGCCGTGCCATA
<i>cat</i>	ABO92401.1	CAATCCTCAATCGACACGAA	GATTGTGTAGCAAGGCAGCA
<i>mdtL</i>	CAR15381.2	CGGACAAACCACGAGAAAAT	GAAGGTGAGGATCACCGAA
<i>mdtF</i>	KEL93478.1	GGACCCGCAAAAACCTCAATA	AGTTGACCACCGGAAATCTG

Antimicrobial resistance gene	Accession number	Forward primer	Reverse primer
<i>ermF</i>	BAD66041.1	AGCACCCGCTTTTTCCTTAT	GATCAAGAGGGGCTTTAGGG
<i>ermB</i>	BAH18720.1	GGTTGCTCTTGACACTCAA	CTGTGGTATGGCGGGTAAGT
<i>ermG</i>	122586.NMB0066	TGCTGTCTTTACAGGCCACT	GCATATGTTCCAGTCCCTTCA
<i>ermC</i>	BAE05991.1	TGAAATCGGCTCAGGAAAAG	GGTCTATTTCAATGGCAGTTACG
<i>mefA_10</i>	583346.CKR_2320	CCTGCAAATGGCGATTATTT	CCAAAGACCGCATAGGGTAA
<i>mefA_3</i>	286636.M6_SPY1166	TTACCCTATGCGGTCTTTGG	GAACCAGCTGCTGCGATAAT
<i>macB</i>	ACR63203.1	GGCTGGAAGACCGTACAGAG	GTTGGTTCATCGGCAAGAAT
<i>fosB</i>	NP_372857.1	AGCTTGCAGGCCTATGGAT	TGCCAATATTTAAATTCGCTGTC
<i>cfr</i>	YP_003896025.1	CAACGAAGGGCAGGTAGAA	GACCACAAGCAGCGTCAATA
<i>mfsA</i>	WP_002584949.1	AATATGCTCTCCGGGCTTTT	TTTGCACACCGTAAAATGGA
<i>ermA</i>	AB047088.2	GAGGGGTTTACCGCTTCTTT	ATCGGATCAGGAAAAGGACA
<i>mecA</i>	YP_184944.1	TCCAGGAATGCAGAAAGACC	GGCCAATTCCACATTGTTTC
<i>arnA</i>	CAR03684.2	GAAATTCACCGTCTGGTCGT	GTGGTGCAACAGAAATCACG
<i>mdtO</i>	ASB78387.1	TTGTTGGCCTCTATCCAACC	TTAAGCGCTTGATGCATTTG
<i>qacA</i>	YP_536864	GACCCTTCTGGTACCCAACA	TCCCCATTTATCAGCAAAGG
<i>qacC</i>	CAA86016.1	TGGGCGGGACTAGGTTTAG	ACGAAACTACGCCGACTATGA
<i>cm1A1</i>	ABI50481.1	AATGGGATGCCTGATAGCTG	ACCCACTAGCCACATTGGAG
<i>qacE</i>	NC_001735.4	TCGGTGTTGCTTATGCAGTC	ATCAAGCTTTTGCCCATGAA
<i>qnrA</i>	ACA43024.1	ATTTCTCAGCCAGGATTG	ACTGCAATCCTCGAACTGG
<i>qnrB</i>	AFD54601.1	CGATCTGACCAATTCGGAGT	ACGATGCCTGGTAGTTGTCC
<i>qnrC</i>	ACK75961.1	GCAGAATTCAGGGGTGTGAT	AACTGCTCCAAAAGCTGCTC
<i>qnrS</i>	AEG74318.1	TGGAAACCTACCGTCACACA	AATCGCATCGGATAAAGGTG
<i>spc</i>	AAL05549.1	TGACGAACGCAATGTGATT	TCAGTGCCAGATCTTTTGA
<i>vatA</i>	AAF24087.1	AACAGCTTCTGCAGCAATGA	CCTTGAAAGGGGACATTGAA
<i>vatB</i>	AAA86871.1	TGGGAAAAAGCAACTCCATC	TTCTGACCAATCCACACATCA
<i>aadE</i>	CAZ55809.1	TGTGCCGCAAAGAGATACTG	AACCTTCCACGACATCATCC
<i>sul1</i>	ADB23338.1	AGGCTGGTGGTTATGCACTC	AAGAACCGCACAATCTCGTC
<i>tetQ</i>	Y08615.1	GCAAAGGAAGGCATACAAGC	AAACGCTCCAAATTCACACC
<i>tetX</i>	ABQ05845.1	CGGTACGCTGGATTACACA	CATCGGAATTGCCTTTTTGT
<i>tetW</i>	ACD97480.1	GGTGCA GTTGGAGTTGTTT	AAATGACGGAGGGTTCCTTT
<i>tetM</i>	ACO22036.1	TTGATGCGGGAAAACTACC	TACCTCTGTCCACGCTTCCT
<i>tetO</i>	EAQ71799.1	GCGTCAAAGGGGAATCACTA	CGGTATACTTCCGCCAAAAA
<i>tetB</i>	AAL09908.1	CAAACTTGCCCCTAACCAA	GCTTTCAGGGATCACAGGAG
<i>dfrA27</i>	ACD45689.1	TTGTTGTTTCTGGTGGTGA	TTCCGGAAAGAAACATTGC
<i>dfrF</i>	AEBU01000146.1	GATTGTTGCGAGGTCAAAGAA	CGCCCCATAATAACCACATT
<i>vanUG</i>	ACR77286.1	ATTTGCGAAACTCGGAAAAA	ACACCTCATTTTCGGGTACG
<i>vanR</i>	CAJ68489.1	TGAAGCTGTATGGGGAGAAAA	TTTCGGGTTTTTAGAAGTTCA
<i>vanA</i>	ACP19236.1	GTGCGGTATTGGGAAACAGT	TGCGTTTTTCAGAGCCTTTTT
<i>vanB</i>	WP_032489746.1	CCTGCCTGGTTTACATCGT	GCTGTCAATCAGTGCAGGAA
<i>vanX</i>	NP_878017.1	CCGTTGACGGTTATGAAGT	CAGCCAGTTCTTTTGCCTTC
<i>cfr_2</i>	AJ249217.1	GCCGGAGCTTTTCTCTACT	GGTGCCGAAAGTCAAAACAT
<i>sulA</i>	AAB03082.1	GATATTCGCGGTTTTCCAGAACC	GTCTTGACCCGAATGCATAAC
16S rRNA	a	CAACGCGARGAACCTTACC	ACAACACGAGCTGACGAC

a The primers for the 16S rRNA gene were previously described by Gloor *et al.* (2010), PLoS ONE 5(10): e15406. doi:10.1371/journal.pone.0015406

27 **Supplementary Table 2.** Results 16S rRNA gene profiling (aggregated to order level). Numbers represent the abundance, as percentage of the  
 28 total microbiome, of each indicated taxa.

	hospital sewage t1	hospital sewage t2	hospital sewage t3	hospital sewage t4	urban WWTP influent t1	urban WWTP influent t2	urban WWTP influent t3	urban WWTP influent t4	suburban WWTP influent t1	suburban WWTP influent t2	suburban WWTP influent t3	suburban WWTP influent t4
Clostridiales	29.50	27.42	27.41	30.42	26.94	13.34	17.65	14.53	18.40	21.10	19.63	16.84
Lactobacillales	15.18	18.49	16.99	9.31	15.23	20.57	8.61	7.43	10.85	9.42	8.69	14.43
Pseudomonadales	8.03	7.01	4.99	7.95	9.21	3.92	5.25	2.05	4.58	4.55	3.82	3.39
Bacteroidales	6.88	7.57	4.35	5.92	4.33	5.78	4.19	2.24	5.36	5.19	7.34	3.65
Burkholderiales	6.87	5.32	9.08	7.47	5.59	3.26	5.35	6.16	4.73	4.94	5.10	4.38
Aeromonadales	4.96	6.70	3.78	4.15	2.16	19.29	11.11	9.91	13.06	7.54	11.43	17.26
Actinomycetales	3.86	2.29	4.04	5.14	5.02	3.42	3.59	3.35	4.12	3.03	3.97	4.67
Campylobacteriales	3.77	6.08	5.63	6.61	4.31	10.06	17.55	29.03	16.95	15.13	18.24	12.36
Bifidobacteriales	3.71	4.36	4.21	4.82	5.34	2.05	3.77	2.57	2.68	2.30	3.06	2.46
Coriobacteriales	2.67	2.50	2.81	2.89	2.85	1.43	2.41	2.15	1.72	1.77	1.98	2.00
Flavobacteriales	1.43	0.90	1.76	1.64	0.76	0.33	0.79	0.46	0.39	0.93	0.37	0.28
Rhodocyclales	1.31	0.89	1.57	1.42	1.36	2.49	2.57	3.54	3.48	3.05	3.91	3.21
Enterobacteriales	1.11	1.24	1.58	1.27	1.41	1.94	2.00	1.45	1.46	1.60	1.56	1.95
Rhodobacterales	0.83	0.37	1.04	0.78	1.04	0.21	0.66	0.66	0.34	0.72	0.28	0.37
Neisseriales	0.73	0.64	0.99	0.72	0.65	0.33	0.70	0.89	0.41	0.64	0.64	0.42
Rhizobiales	0.52	0.39	0.70	0.60	0.64	0.17	0.47	0.54	0.23	0.59	0.20	0.24
Xanthomonadales	0.50	0.22	0.60	0.50	0.33	0.09	0.26	0.37	0.12	0.30	0.13	0.12
Fusobacteriales	0.48	0.14	0.12	0.31	1.07	1.99	1.79	0.52	1.82	2.49	1.08	2.08
Erysipelotrichales	0.47	0.49	0.89	0.61	0.59	0.12	0.21	0.30	0.38	0.23	0.21	0.21
Desulfovibrionales	0.32	0.28	0.38	0.37	0.29	0.59	0.39	0.24	0.61	0.36	0.81	0.67
ASSO-13- (Rhodocyclales)	0.32	0.26	0.49	0.22	0.29	0.07	0.30	0.48	0.15	0.29	0.13	0.12
Synergistales	0.27	0.09	0.05	0.16	0.98	0.79	0.98	0.98	0.92	2.15	1.24	1.25
Unassigned	3.77	4.85	3.95	4.27	4.03	5.56	4.42	4.77	4.91	4.29	3.82	5.14
others	2.48	1.48	2.58	2.45	5.55	2.20	4.96	5.36	2.31	7.39	2.37	2.48

	urban effluent t1	urban effluent t2	urban effluent t3	urban effluent t4	river water site 1 t1	river water site 1 t2	river water site 1 t3	river water site 1 t4	river water site 2 t1	river water site 2 t2	river water site 2 t3	river water site 2 t4
Clostridiales	9.83	10.70	7.24	11.31	6.28	3.75	6.13	5.94	6.65	8.45	7.78	9.21
Lactobacillales	1.42	2.85	1.06	2.09	1.10	0.97	1.04	1.22	1.32	0.94	1.58	1.55
Pseudomonadales	0.89	2.72	0.60	0.79	0.37	0.57	0.32	0.46	0.66	0.90	0.83	1.04
Bacteroidales	1.95	1.73	1.12	2.11	0.66	0.83	1.11	1.40	1.25	1.25	1.67	2.35
Burkholderiales	12.56	13.08	10.98	8.45	14.40	11.96	8.17	8.32	9.41	12.89	11.97	7.66
Aeromonadales	1.17	1.98	0.65	1.23	0.43	0.28	0.58	1.07	0.77	0.67	1.48	1.69
Actinomycetales	12.08	23.05	18.25	19.38	13.50	11.93	17.20	17.91	17.09	16.85	14.27	17.41
Campylobacteriales	2.71	2.63	1.28	3.46	1.63	0.97	0.93	1.41	0.92	1.91	3.15	3.27
Bifidobacteriales	0.61	0.47	0.46	0.83	0.58	0.37	0.43	0.58	0.55	0.48	0.54	0.77
Coriobacteriales	0.67	1.00	0.53	0.93	0.41	0.30	0.47	0.43	0.54	0.53	0.84	0.84
Flavobacteriales	0.44	10.96	0.46	0.40	0.38	0.56	0.53	0.58	0.42	0.41	0.50	0.49
Rhodocyclales	7.46	4.69	9.52	8.69	13.89	16.47	12.32	13.90	9.49	7.81	9.79	8.48
Enterobacteriales	0.46	0.59	0.30	0.89	0.18	0.13	0.18	0.29	0.38	0.31	0.40	0.68
Rhodobacterales	0.56	0.32	0.96	0.80	0.87	1.56	1.48	1.60	1.42	0.79	0.96	0.95
Neisseriales	1.97	0.18	1.32	0.78	1.04	0.82	0.49	0.41	0.75	1.89	1.33	0.55
Rhizobiales	1.05	0.44	1.40	1.28	1.31	2.04	1.96	2.26	1.76	1.14	1.39	1.47
Xanthomonadales	0.30	0.28	0.56	0.39	0.72	0.86	0.71	0.82	0.47	1.37	0.54	0.50
Fusobacteriales	0.22	0.16	0.12	0.06	0.02	0.05	0.13	0.07	0.08	0.11	0.13	0.08
Erysipelotrichales	0.08	0.08	0.03	0.11	0.03	0.04	0.05	0.05	0.06	0.05	0.10	0.12
Desulfovibrionales	0.10	0.20	0.11	0.18	0.06	0.03	0.06	0.08	0.09	0.09	0.15	0.13
ASSO-13 (Rhodocyclales)	0.02	0.01	0.01	0.03	0.05	0.04	0.03	0.06	0.03	0.03	0.04	0.07
Synergistales	0.22	0.22	0.34	0.36	0.15	0.05	0.33	0.18	0.22	0.20	0.10	0.32
Unassigned	3.97	3.59	3.59	3.65	1.88	3.32	3.03	2.94	3.68	3.72	3.51	2.90
others	39.26	18.05	39.10	31.80	40.05	42.08	42.30	38.01	41.99	37.20	36.93	37.46





30 **Supplementary Table 3:** Aggregated cumulative abundance of ARG classes relative to the 16S rRNA gene.

	aminoglycosides	bacitracin	$\beta$ -lactams	chloramphenicols	efflux	macrolides	polymyxins
hospital sewage t1	1429.19	11.30	513.11	20.76	47.37	361.11	2.11
hospital sewage t2	1423.39	12.57	568.49	20.60	47.32	375.27	2.15
hospital sewage t3	1660.78	12.89	605.79	16.78	51.58	403.89	2.15
hospital sewage t4	1633.67	12.06	590.51	20.00	46.05	358.19	1.75
urban WWTP influent t1	87.02	5.83	38.72	9.31	27.77	163.60	0.92
urban WWTP influent t2	247.49	4.23	27.49	14.39	15.23	235.73	0.86
urban WWTP influent t3	106.03	3.46	41.34	15.22	21.21	107.09	0.92
urban WWTP influent t4	159.04	4.49	45.60	20.35	25.42	134.69	0.96
suburban WWTP influent t1	124.07	3.30	30.57	10.86	18.79	64.30	0.70
suburban WWTP influent t2	119.86	7.26	43.60	13.32	23.07	135.36	0.86
suburban WWTP influent t3	118.46	5.28	37.64	9.23	18.83	164.84	0.77
suburban WWTP influent t4	193.30	3.02	32.04	15.53	21.74	232.83	1.07
urban WWTP effluent t1	34.35	2.74	14.75	1.95	4.60	20.79	0.24
urban WWTP effluent t2	36.57	1.48	10.30	1.88	3.72	24.04	0.27
urban WWTP effluent t3	25.47	1.63	10.03	1.48	2.69	11.21	ND
urban WWTP effluent t4	43.17	5.00	23.40	3.43	7.61	27.53	0.40
river water site 1 t1	29.28	2.37	10.27	1.47	3.90	22.08	0.23
river water site 1 t2	25.94	0.97	4.82	0.38	1.64	15.71	ND
river water site 1 t3	26.28	1.39	7.29	0.98	2.48	14.97	ND
river water site 1 t4	30.90	1.84	10.63	1.74	3.47	19.36	ND
river water site 2 t1	18.36	2.30	9.70	1.37	3.39	13.78	ND
river water site 2 t2	39.02	2.35	10.50	1.41	3.23	19.80	0.20
river water site 2 t3	39.99	2.57	14.78	2.04	4.62	19.37	ND
river water site 2 t4	58.06	4.57	26.27	4.12	7.35	31.70	0.42

31

32

	QAC resistance genes	quinolones	streptogramins	sulphonamides	tetracyclines	trimethoprim	vancomycin
hospital sewage t1	344.47	9.17	ND	216.50	384.68	12.17	386.38
hospital sewage t2	337.67	6.91	0.07	214.56	387.78	14.31	302.88
hospital sewage t3	388.42	7.15	ND	234.68	402.38	11.04	377.20
hospital sewage t4	371.23	6.61	ND	243.17	390.20	15.34	457.12
urban WWTP influent t1	45.50	5.58	1.38	29.45	238.76	10.80	2.03
urban WWTP influent t2	93.93	6.87	0.90	59.21	197.41	2.41	ND
urban WWTP influent t3	82.65	4.96	1.34	54.48	186.89	5.54	1.81
urban WWTP influent t4	88.49	7.14	1.43	59.19	180.66	6.47	3.63
suburban WWTP influent t1	10.83	7.56	1.11	8.26	86.37	4.59	0.00
suburban WWTP influent t2	68.33	6.05	1.62	45.47	215.08	4.66	3.30
suburban WWTP influent t3	77.72	4.90	1.10	47.07	209.39	3.82	0.29
suburban WWTP influent t4	93.17	5.59	1.30	58.94	189.38	3.93	0.18
urban WWTP effluent t1	60.35	0.91	0.00	38.49	45.90	1.43	0.00
urban WWTP effluent t2	25.52	0.81	0.00	17.44	47.16	0.77	1.31
urban WWTP effluent t3	70.48	0.45	0.00	43.60	30.13	0.79	0.00
urban WWTP effluent t4	76.09	2.39	0.00	54.03	70.46	1.79	0.00
river water site 1 t1	53.19	0.80	ND	35.46	39.29	1.17	ND
river water site 1 t2	61.59	0.32	ND	43.29	23.08	0.68	0.25
river water site 1 t3	60.13	0.33	ND	49.54	30.12	0.92	ND
river water site 1 t4	67.05	0.81	ND	49.71	35.37	0.96	ND
river water site 2 t1	56.33	0.57	ND	42.26	33.41	0.79	ND
river water site 2 t2	71.04	0.76	ND	45.97	42.81	0.87	0.76
river water site 2 t3	68.26	0.74	ND	52.54	45.71	1.25	ND
river water site 2 t4	90.82	1.82	ND	59.42	63.83	2.15	1.49

33

34 Abundance for each individual ARG was calculated relative to the CT-value of 16S rRNA in each sample, as described in the Materials and  
35 Methods. Abundances of each ARG detected per ARG class were then aggregated to determine the relative abundance of ARG classes. ND:  
36 not detectable.

